PATENT COOPERATION TREATY

PCT

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

(Chapter II of the Patent Cooperation Treaty)

(PCT Article 36 and Rule 70)

Applicant's or agent's file reference 27558	FOR FURTHER ACTION	See Form PCT/IPEA/416
International application No. PCT/IL04/00181	International filing date *(day/month/year)* 24 February 2004 (24.02.2004)	Priority date *(day/month/year)* 27 April 2003 (27.04.2003)
International Patent Classification (IPC) or national classification and IPC IPC: C12P 21/06; C12N 9/00, 9/14, 1/12, 1/20, 5/00, 15/00; C07H 21/04; A01H 11/00 USPC: 435/4, 6, 41, 69.1, 183, 195, 252.1, 252.3, 254.1, 320.1, 325, 410,; 536/23.1, 23.4, 23.5; 800/295		
Applicant METABOGAL, LTD		

1. This report is the international preliminary examination report, established by this International Preliminary Examining Authority under Article 35 and transmitted to the applicant according to Article 36.
2. This REPORT consists of a total of ___ sheets, including this cover sheet.
3. This report is also accompanied by ANNEXES, comprising:
   a. ☐ *(sent to the applicant and to the International Bureau)* a total of ___ sheets, as follows:
      ☐ sheets of the description, claims and/or drawings which have been amended and are the basis of this report and/or sheets containing rectifications authorized by this Authority (see Rule 70.16 and Section 607 of the Administrative Instructions).
      ☐ sheets which supersede earlier sheets, but which this Authority considers contain an amendment that goes beyond the disclosure in the international application as filed, as indicated in item 4 of Box No. I and the Supplemental Box.
   b. ☐ *(sent to the International Bureau only)* a total of (indicate type and number of electronic carrier(s)) _____ , containing a sequence listing and/or tables related thereto, in electronic form only, as indicated in the Supplemental Box Relating to Sequence Listing (see Section 802 of the Administrative Instructions).
4. This report contains indications relating to the following items:

☒	Box No. I	Basis of the report
☐	Box No. II	Priority
☐	Box No. III	Non-establishment of opinion with regard to novelty, inventive step and industrial applicability
☒	Box No. IV	Lack of unity of invention
☒	Box No. V	Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement
☐	Box No. VI	Certain documents cited
☐	Box No. VII	Certain defects in the international application
☐	Box No. VIII	Certain observations on the international application

Date of submission of the demand	Date of completion of this report
29 November 2004 (29.11.2004)	30 March 2007 (30.03.2007)
Name and mailing address of the IPEA/ US Mail Stop PCT, Attn: IPEA/US Commissioner for Patents P.O. Box 1450 Alexandria, Virginia 22313-1450 Facsimile No. (571) 273-3201	Authorized officer Manjunath N. Rao, Ph.D. Telephone No. 571-272-1600

MANJUNATH N. RAO, PH.D.
PRIMARY EXAMINER

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International application No.
PCT/IL04/00181

Box No. I Basis of the report

1. With regard to the **language**, this report is based on:

☒ the international application in the language in which it was filed.

☐ a translation of the international application into _____, which is the language of a translation furnished for the purposes of:

- ☐ international search (under Rules 12.3 and 23.1(b))
- ☐ publication of the international application (under Rule 12.4(a))
- ☐ international preliminary examination (under Rules 55.2(a) and/or 55.3(a))

2. With regard to the **elements** of the international application, this report is based on *(replacement sheets which have been furnished to the receiving Office in response to an invitation under Article 14 are referred to in this report as "originally filed" and are not annexed to this report)*:

☒ the international application as originally filed/furnished

☒ the description:
pages 1-75 as originally filed/furnished
pages* NONE received by this Authority on _____
pages* NONE received by this Authority on _____

☒ the claims:
pages 76-84 as originally filed/furnished
pages* NONE as amended (together with any statement) under Article 19
pages* NONE received by this Authority on _____
pages* NONE received by this Authority on _____

☒ the drawings:
pages 1-17 as originally filed/furnished
pages* NONE received by this Authority on _____
pages* NONE received by this Authority on _____

☒ a sequence listing and/or any related table(s) - see Supplemental Box Relating to Sequence Listing.

3. ☐ The amendments have resulted in the cancellation of:

- ☐ the description, pages _____
- ☐ the claims, Nos. _____
- ☐ the drawings, sheets/figs _____
- ☐ the sequence listing (*specify*): _____
- ☐ any table(s) related to the sequence listing (*specify*): _____

4. ☐ This report has been established as if (some of) the amendments annexed to this report and listed below had not been made, since they have been considered to go beyond the disclosure as filed, as indicated in the Supplemental Box (Rule 70.2(c)).

- ☐ the description, pages _____
- ☐ the claims, Nos. _____
- ☐ the drawings, sheets/figs _____
- ☐ the sequence listing (*specify*): _____
- ☐ any table(s) related to the sequence listing (*specify*): _____

** If item 4 applies, some or all of those sheets may be marked "superseded."*

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International a

PCT/IL04/00181

Box No. IV Lack of unity of invention

1. ☒ In response to the invitation to restrict or pay additional fees the applicant has, within the applicable time limit:

   ☐ restricted the claims.

   ☒ paid additional fees.

   ☐ paid additional fees under protest, and, where applicable, the protest fee

   ☐ paid additional fees under protest but the applicable protest fee was not paid

   ☐ neither restricted the claims nor paid additional fees

2. ☐ This Authority found that the requirement of unity of invention is not complied with and chose, according to Rule 68.1, not to invite the applicant to restrict or pay additional fees.

3. This Authority considers that the requirement of unity of invention in accordance with Rules 13.1, 13.2 and 13.3 is:

   ☐ complied with.

   ☒ not complied with for the following reasons:

4. Consequently, this report has been established in respect of the following parts of the international application:

   ☐ all parts

   ☒ the parts relating to claims Nos. 1-24,28-31,33-37 and 42

INTERNATIONAL PRELIMINARY REPORT ON PATENTABILITY

International PCT/IL04/00181

Box No. V Reasoned statement under Article 35(2) with regard to novelty, inventive step or industrial applicability; citations and explanations supporting such statement

1. Statement

Novelty (N)	Claims 4, 12-24, 42	YES
	Claims 1-3, 5-11, 28-31, 33-37	NO
Inventive Step (IS)	Claims NONE	YES
	Claims 1-24, 28-31, 33-37, 42	NO
Industrial Applicability (IA)	Claims 1-24, 28-31, 33-37, 42	YES
	Claims NONE	NO

2. Citations and Explanations (Rule 70.7)
Please See Continuation Sheet

Supplemental Box Relating to Sequence Listing

Continuation of Box No. I, item 2:

I. With regard to any nucleotide and/or amino acid sequence disclosed in the international application and necessary to the claimed invention, this report was established on the basis of:

a. type of material

☒ a sequence listing

☐ table(s) related to the sequence listing

b. format of material

☒ on paper

☒ in electronic form

c. time of filing/furnishing

☒ contained in the international application as filed

☒ filed together with the international application in electronic form

☐ furnished subsequently to this Authority for the purposes of search and/or examination

☐ received by this Authority as an amendment* on _____

2. ☐ In addition, in the case that more than one version or copy of a sequence listing and/or table(s) relating thereto has been filed or furnished, the required statements that the information in the subsequent or additional copies is identical to that in the application as filed or does not go beyond the application as filed, as appropriate, were furnished.

3. Additional comments:

* *If item 4 in Box No. I applies, the listing and/or table(s) related thereto, which form part of the basis of the report, may be marked "superseded."*

Supplemental Box

In case the space in any of the preceding boxes is not sufficient.

Continuation of:

V. 2. Citations and Explanations:

Claims 1-3, 5-11, 28-31, 33-37 lack novelty under PCT Article 33(2) as being anticipated by Martin et al. (DNA, 1988, Vol. 7, No.2, pages 99-106). Claims 1-3, 5-11, 28-31, 33-37 are drawn to a host cell producing a high mannose recombinant protein comprising a polynucleotide encoding the recombinant protein and a signal for causing the recombinant protein to be produced as a high mannose protein, wherein the polynucleotide comprises a first nucleic acid sequence encoding said protein of interest operably linked to a second nucleic acid sequence encoding a signal peptide wherein said signal peptide comprises a ER targeting peptide and wherein said host cell is a prokaryotic or a eukaryotic host cell and wherein said polypeptide is one of the lysosomal proteins such as glucocerebrosidase. Claim 28-31, 33-37 are also drawn to a recombinant biologically active high mannose lysosomal enzyme having at least one oligosaccharide chain comprising an exposed mannose residue. Martin et al. disclose one such host cell comprising a polynucleotide encoding said enzyme wherein said polypeptide is produced as a high-mannose protein in high levels. Matin et al. also disclose a recombinant glucocerebrosidase wherein said enzyme is inherently a biologically active high mannose lysosomal enzyme having at least one oligosaccharide chain comprising an exposed mannose residue. Thus, Martin et al. anticipate claims 1-3, 5-11, 28-31, 33-37 as written.

Claims 4, 12-24 and 42 lack an inventive step under PCT Article 33(3) as being obvious over the prior art as applied in the immediately preceding paragraph and further in view of Boller et al. and Zhu et al. Claims 4, 12-24 and 42 are drawn to a host cell producing a high mannose recombinant protein comprising a polynucleotide encoding the recombinant protein and a signal for causing the recombinant protein to be produced as a high mannose protein, wherein the polynucleotide comprises a first nucleic acid sequence encoding said protein of interest operably linked to a second nucleic acid sequence with with SEQ ID NO:1 encoding a signal peptide wherein said signal peptide comprises a ER targeting peptide and wherein said polynucleotide is operably linked to a third polynucleotide sequence with SEQ ID NO:2 encoding a plant vacuolar targeting sequence, and wherein said host cell is a plant cell and wherein said polypeptide

Supplemental Box

is one of the lysosomal proteins such as glucocerebrosidase. Claim 42 is drawn to a recombinant protein produced from a plant host cell. The reference of Martin et al. has already been discussed above. Martin et al. teach the production of glucocerebrosidase, a lysosomal protien recombinantly using a host cell comprising a polynucleotide with a signal sequence. The reference of Zhu et al. teach the polynucleotide encoding the signal peptide SEQ ID NO:1 and its use in producing novel recombinant proteins. On similar lines Boller et al. teach the vacuolar targeting sequence SEQ ID NO:2 and its use in targeting polypeptides into the vacuolar space. The invention as a whole is directed to production of glucocerebrosidase as a transgenic protein in plant host cells. The art and the above references teach and provide all sequences required for expressing the glucocerebrosidase as a transgenic protein. The production of mammalian proteins in plant products such as fruits and seed is well known since it eliminates the steps of purification and makes the recombinant protein ready for administration as a plant product. Therefore, with the above references in hand, it would have been obvious to one of ordinary skill in the art to produce human glucocerebrosidase, which is used in enzyme replacement therapy for lysosomal enzyme disorders, as a plant protein by expressing as a polynucleotide linked to the above signal sequence and vacuolar targeting sequences. One of ordinary skill in the art would have been motivated to do so since the lysosomal protein is extensively used in enzyme replacement therapy and production of the protein as a plant product would avoid the extensive purification steps and can be easily administered as a plant product. One of ordinary skill in the art would have had a reasonable expectation of success since Martin et al. already provide a host cell producing the high-mannose protein, Zhu et al. and Boller et al. provide the sequences to make a DNA construct to be expressed in a plant cell. Therefore the above invention would have been *prima facie* obvious to one of ordinary skill in the art.

Claims 1-24, 28-31, 33-37, 42 meet the criteria set out in PCT Article 33(4), and thus have industrial applicability because the subject matter claimed can be made or used in industry.